World Journal of Microbiology & Biotechnology **16**: 507–512, 2000.

Fermentative production of acetic acid from various pure and natural cellulosic materials by *Clostridium lentocellum* SG6

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Received 25 October 1999; accepted 12 May 2000

Keywords: Acetic acid production, agricultural residues, anaerobic fermentation, biomass to acetic acid, direct microbial conversion of cellulose

Summary

Clostridium lentocellum SG6 fermented various pure crystalline cellulosic materials efficiently with maximum acetic acid yield (gram acetic acid/gram substrate) of 0.67, at low substrate (8 g l^{-1}) concentration. The strain grew poorly on crude biopolymers but fermented them easily after alkali treatment, when grown with 8 g substrate l^{-1} concentration of alkali-extracted cotton straw (AECS), paddy straw (AEPS) and sorghum stover (AESS) etc. The acetic acid to substrate (A/S) ratios were similar to those obtained with pure cellulosic materials. An increase in substrate concentration led to a decreased A/S ratio and a decreased percentage of substrate degraded. At high substrate concentration of 75 g filter paper l^{-1}, the strain SG6 converted 63.2 g filter paper into 31.28 g acetic acid l^{-1}. At 100 g l^{-1} concentrations, AECS and AEPS served as the best substrates for acetic acid production when compared with other biopolymers. A maximum amount of 30.98 and 30.86 g acetic acid was produced from 70.6 g AEPS and 70.1 g AESS l^{-1} of medium by strain SG6, respectively. Acetic acid production of 0.67 g g^{-1} pure cellulose (Whatman No. 1 filter paper), 0.63 g g^{-1} of alkali-treated cotton straw (AECS) are the highest among the cellulolytic bacteria reported so far in mono culture fermentations with pure and native cellulosic materials.

Introduction

Acetic acid is widely used in the food, pharmaceutical and textile industries and is also an important component in the synthesis of many chemicals, such as vinyl acetate, cellulose acetate, acetic acid esters, terepthalic acid etc. (Ghose & Bhadra 1985; Cheryan *et al*. 1997). The microbial conversion of abundant and renewable cellulosic biomass to organic acids and ethanol offers an attractive alternative to fuels and basic chemical feed stocks derived from petroleum (Ghose & Bhadra 1985; Lowe *et al*. 1993; Beguin & Aubert 1994; Lee 1997). Cellulosic biomass is available in enormous quantities as waste of agricultural, industrial, and forestry residues, municipal solids waste etc. The microbial conversion of these wastes to generate value-added products can also alleviate pollution problems.

The direct conversion of cellulosic biomass to acetic acid by single step fermentation can be economical over the expensive multi-step process in which fungal cellulases, yeasts and acetogenic bacteria are used. (Lynd 1989; Parisi 1989; Ebner *et al*. 1996). Moreover, 3 M of acetic acid are produced by anaerobic fermentations from 1 M of glucose equivalents fermented, whereas in aerobic breakdown only 2 M of acetic acid are produced (Ghose & Bhadra 1985; Sugaya *et al*. 1986; Cheryan *et al*. 1997). In this direction, we have reported the direct conversion of biomass to acetic acid by various cellulolytic, mesophilic and anaerobic *Clostridium* spp. (Ravinder *et al*. 1998). One of these strains, identified as *C. lentocellum* SG6, produced high amounts of acetic acid, 0.63 g g^{-1} of cellulose (Whatman No. 1 filter paper).

An agrarian country such as India generates about 388 million tonnes of biomass per annum, which includes paddy straw, cotton straw, sorghum stover, grass etc. (Anonymous 1991). In order to develop a practical process for acetic acid production, it is necessary to evaluate the ability of the strain to degrade natural cellulosic substrates. In the present study, the ability of *C. lentocellum* SG6 to ferment high concentrations of various pure cellulosic materials and pre-treated agricultural materials such as paddy straw, cotton straw, sorghum stover, grass etc., to produce acetic acid is reported.

Materials and Methods

Fermentation experiments

Clostridium lentocellum SG6 was grown in 120 ml serum vials with 20 ml of pre-reduced CMS medium (Sai Ram

& Seenayya 1991) containing (g l^{-1}); KH_2PO_4, 1.5; K_2HPO_4, 2.0; urea, 2.0; $MgSO_4$, 0.8; $CaCl_2$, 0.15; sodium citrate, 3.5; cysteine HCl, 0.15; yeast extract, 5.0; resazurin, 0.002; cellulose, 8.0; in N_2 atmosphere. The medium was sterilized by autoclaving at 121 °C for 30 min. The pH of the fermentation medium was adjusted to 7.2 before inoculation. A 5% (v/v) inoculum, grown on 4 g cellulose l^{-1}, was added and incubations were carried out at 37 ± 2 °C without shaking. The presence of growth was assessed by visual observation and substrate degradation. The medium was buffered with 0.5, 1.24, 2.5, 3.74, 5.0 and 6.24% $CaCO_3$ at 10, 25, 50, 75, 100 and 125 g cellulosic substrate l^{-1}, respectively. Triple strength CMS medium was used when the substrate concentration was above 10 g l^{-1}. The carbon sources used were cellulose (Whatman No. 1 filter paper), tissue paper, Avicel, Solka Floc, native cotton, carboxymethylcellulose sodium salt (low viscosity, Sigma) and the agricultural materials, cotton straw, paddy straw, grass, ground nut shells, sorghum stover, corn cobs and castor straw.

Preparation of the treated biopolymers

The agricultural materials were cut into pieces of approximately 1 cm, ground with a mortar and pestle and extracted by boiling with distilled water for 30 min. Alkali-extracted fractions were prepared by autoclaving the agricultural materials at 121 °C for 15 min with 1% (w/v) NaOH, followed by neutralization with H_2SO_4. These fractions were thoroughly washed with distilled water and dried at 60 °C for 48 h (Sai Ram & Seenayya 1991).

Estimations

Undegraded cellulose was estimated gravimetrically by the method of Weimer & Zeikus (1977). Cultures were centrifuged at 25,000 × *g* for 15 min and the supernatant was drawn off gently with a Pasteur pipette. The pellet was resuspended in 8% formic acid to lyse the cells. The process was repeated to eliminate residual $CaCO_3$ if any. This solution was then passed through preweighed 0.45 μ Millipore filters. The filters were dried at 60 °C to constant weight and the residual cellulose was determined by difference.

Residual carboxymethylcellulose sodium salt (NaCMC) in fermented broth was determined after acid hydrolysis by the DNS method. 50 μl of 5 M H_2SO_4 solution was added to 0.5 ml of fermented broth samples. These samples were placed in a steam bath for 3 h and then neutralized by the addition of 35 μl of 10 N NaOH solution. The reducing sugars released were determined by the DNS method. The amount of undegraded NaCMC was estimated by finding reducing sugar values from the calibration curve of standard 1% NaCMC solution, which was treated by the same procedure as described above.

Acetic acid and ethanol were determined by gas chromatography of a sample of centrifuged fermentation broth acidified with H_3PO_4 (Swamy & Seenayya 1996).

The results reported are the arithmetic mean values of three experiments in triplicate carried out on different occasions. Appropriate tests of significance – analysis of variance (one way and two way ANOVA with interactions). *F*-test and C.D at 5% were utilized (Visweswara Rao 1996) and the results are provided in the text.

Results

Fermentation of different pure cellulosic materials

Clostridium lentocellum SG6 efficiently fermented pure crystalline cellulosic materials such as filter paper (FP), tissue paper (TP), microcrystalline Avicel, and native cotton, which efficiently resulted in more than 90% degradation of the initial cellulose at 8 g l^{-1} concentration (Table 1). Maximum amounts of acetic acid and acetic acid/ethanol (A/E) ratio were obtained on FP and Avicel. The next best substrates were Solka Floc (SF), followed by TP and native cotton. The strain grew sluggishly on the sodium salt of caroboxymethylcellulose and only degraded 57% of the substrate. The production

Table 1. Fermentation of various pure cellulosic materials by *C. lentocellum* SG6.

Substrate	Acetic acid (g l^{-1})	Ethanol (g l^{-1})	Substrate degraded (g l^{-1})	Acetate yield (g g^{-1})	A/E ratio
Filter paper	4.91^{a}	1.12^{a}	7.3^{a}	0.67^{a}	$4.38^{a,c}$
Tissue paper	4.48^{b}	1.06^{b}	7.4^{b}	0.60^{b}	$4.23^{b,d,c}$
Avicel	4.82^{c}	1.08^{b}	7.4^{b}	0.65^{c}	4.46^{a}
Solka Floc	4.58^{d}	1.06^{b}	7.3^{a}	0.63^{d}	4.32^{c}
Native cotton	3.91^{e}	0.92^{c}	7.2^{c}	0.54^{e}	4.25^{d}
Carbonxymethylcellulose	2.10^{f}	0.72^{d}	4.9^{d}	0.43^{f}	2.92^{e}
C.D. at 5%	0.045	0.032	0.089	0.005	0.110
F-value	4517.00*	182.83*	1001.98*	2302.54*	229.1*

* $P < 0.0001$.

The values are the average of three experiments, each in triplicate.

Different superscripts indicate that they are significantly different from one another at $P < 0.05$ level.

Initial substrate concentration: 8 g l^{-1}; Incubation time: 5 days.

g g^{-1}: gram acetic acid per gram substrate; A/E: Acetic acid to ethanol ratio.

of acetic acid from these pure cellulosic substrates is significantly different from one another (Table 1).

Fermentation of treated agricultural materials

Water-extracted agricultural materials supported appreciable growth of the strain and also fermentation of substrates (Table 2). Growth was initiated after a lag of 24 h on water-extracted cotton straw, paddy straw, grass, sorghum stover and after 36 h on water-extracted groundnut shells, corn cobs, parthenium weed and castor straw. The strain degraded more than 50% of all water-extracted substrates. The acetic acid production among the water-extracted materials was significant, except for grass and sorghum stover (Table 2). Alkali treatment (delignification) of these agricultural materials further enhanced the utilization of substrate and acetic acid production by the strain SG6 (Table 3). More than 60% of acetic acid yield was obtained on alkali-extracted cotton straw (AECS), paddy straw (AEPS) and sorghum stover (AESS). Acetic acid and ethanol productions by this strain with alkali-treated agricultural materials were similar to those obtained on filter paper (Tables 1 & 3). The fermentation of alkali-extracted agricultural materials by *C. lentocellum* SG6 significantly differed one another in the production of acetic acid, except groundnut shells and corn cobs (Table 3).

Fermentation of crude agricultural materials

The strain SG6 displayed very poor growth on untreated agricultural materials. At 8 g l^{-1} concentration tested, the substrate utilization was less than 10% even after 5 days of incubation and acetic acid was hardly detected.

Acetic acid production at higher concentrations of pure cellulosic (Whatman No.1 filter paper) substrate

The strain grew efficiently at all the cellulose concentrations tested, but displayed differences in the yields of end products, accumulation of reducing sugars and substrate utilization (Table 4). An increase in substrate concentration generally increased the time for cellulose degra-

Table 2. Fermentation of various water extracted agricultural materials by *C. lentocellum* SG6.

Substrate	Acetic acid (g l^{-1})	Ethanol (g l^{-1})	Substrate degraded (g l^{-1})	Acetate yield (g g^{-1})	A/E ratio
Cotton straw	2.36^{a}	0.75^{a}	5.3^{a}	0.44^{a}	3.15^{a}
Paddy straw	2.26^{b}	$0.70^{b,d}$	5.2^{a}	0.43^{b}	3.23^{a}
Grass	1.93^{c}	0.68^{b}	4.8^{b}	0.40^{c}	2.84^{b}
Ground nut shells	1.62^{d}	0.62^{c}	4.7^{b}	0.34^{d}	$2.61^{c,d}$
Sorghum stover	1.92^{c}	0.72^{d}	5.0^{c}	0.38^{e}	2.67^{c}
Corn cobs	1.42^{e}	0.56^{e}	4.3^{d}	0.33^{f}	2.54^{d}
Parthenium weed	1.31^{f}	0.54^{e}	4.2^{d}	0.31^{g}	2.42^{e}
Castor straw	1.36^{g}	0.56^{e}	4.2^{d}	0.32^{h}	2.43^{e}
C.D. at 5%	0.032	0.026	0.139	0.009	0.095
F-value	1272.74*	77.77*	80.84*	229.08*	85.17*

* $P < 0.0001$.
The values are the average of three experiments, each in triplicate.
Different superscripts indicate that they are significantly different from one another at $P < 0.05$ level.
Initial substrate concentration: 8 g l^{-1}; Incubation time: 5 days.
g g^{-1}: gram acetic acid per gram substrate; A/E: Acetic acid to ethanol ratio.

Table 3. Fermentation of various alkali-extracted agricultural materials by *C. lentocellum* SG6.

Substrate	Acetic acid (g l^{-1})	Ethanol (g l^{-1})	Substrate degraded (g l^{-1})	Acetate yield (g g^{-1})	A/E ratio
Cotton straw	4.54^{a}	$1.07^{a,c,d}$	7.2^{a}	0.63^{a}	4.24^{a}
Paddy straw	4.42^{b}	$1.04^{b,c}$	7.2^{a}	0.61^{b}	4.25^{a}
Grass	4.13^{c}	1.02^{b}	7.1^{ab}	0.58^{c}	4.05^{b}
Ground nut shells	4.08^{d}	1.05^{c}	$7.0^{b,c}$	0.58^{c}	3.89^{c}
Sorghum stover	4.34^{e}	$1.09^{d,e}$	7.2^{a}	0.60^{d}	3.98^{b}
Corn cobs	4.05^{d}	$1.11^{e,f}$	$7.1^{a,b}$	0.57^{e}	3.65^{d}
Parthenium weed	3.81^{f}	1.12^{f}	$6.9^{c,d}$	0.55^{f}	3.40^{e}
Castor straw	3.62^{g}	1.06^{e}	6.8^{d}	0.53^{g}	3.42^{e}
C.D. at 5%	0.045	0.025	0.121	0.008	0.081
F-value	369.80*	14.07*	9.97*	145.42*	132.47*

* $P < 0.0001$.
The values are the average of three experiments, each in triplicate.
Different superscripts indicate that they are significantly different from one another at $P < 0.05$ level.
Initial substrate concentration: 8 g l^{-1}; Incubation time: 5 days.
g g^{-1}: gram acetic acid per gram substrate; A/E: Acetic acid to ethanol ratio.

Table 4. Fermentation of various concentrations of cellulose (Whatman No.1 filter paper) by *C. lentocellum* SG6.

Substrate concentration (g l^{-1})	Acetic acid (g l^{-1})	Ethanol (g l^{-1})	Reducing sugars (g l^{-1})	Substrate degraded (g l^{-1})	Acetate yield (g l^{-1})	A/E ratio
10^{x}	5.92^{a}	1.41^{a}	0.12^{a}	9.1^{a}	0.65^{a}	4.20^{a}
25^{x}	14.63^{b}	3.46^{b}	0.34^{b}	23.8^{b}	0.61^{b}	4.23^{a}
50^{xx}	23.10^{c}	5.62^{c}	0.86^{c}	43.2^{c}	0.53^{c}	4.11^{a}
75^{xxx}	31.28^{d}	7.91^{d}	1.33^{d}	63.2^{d}	0.49^{d}	3.95^{b}
100^{xxxx}	31.02^{d}	8.13^{e}	1.71^{e}	72.4^{e}	0.43^{e}	3.81^{c}
125^{xxxx}	30.92^{d}	8.83^{f}	1.92^{f}	78.3^{f}	0.39^{f}	3.50^{d}
C.D. at 5%	0.618	0.094	0.029	0.638	0.016	0.135
F-value	2376.46*	8150.89*	5332.72*	15511.10*	336.88*	38.49*

* $P < 0.0001$.
The values are the average of three experiments, each in triplicate.
Different superscripts indicate that they are significantly different from one another at $P < 0.05$ level.
^{x} Incubated for 5 days; ^{xx} incubated 9 days; ^{xxx} incubated for 12 days; ^{xxxx} incubated for 14 days.
g g^{-1}: gram acetic acid per gram substrate; A/E: Acetic acid to ethanol ratio.
$CaCO_3$ was used at 50% of the substrate concentration as buffering agent.

dation and decreased the acetate yield and the percentage of substrate degraded. *Clostridium lentocellum* SG6 produced acetic acid as the major fermentation product at all the substrate concentrations tested. At up to 25 g cellulose l^{-1}, the concentration of degraded cellulose and end product yields were similar. However, at 50 g cellulose l^{-1} and above, a decrease in acetic acid yield with an increase of ethanol and reducing sugar formation was observed. A maximum amount of 31.28 g acetic acid l^{-1} was obtained from 63.2 g cellulose l^{-1} degraded at a concentration of 75 g cellulose l^{-1}. The acetic acid production by this strain SG6 significantly differed up to 75 g cellulose l^{-1}. After that there was no significant difference up to 125 g cellulose l^{-1} (Table 4).

Fermentation with higher concentrations of alkali-extracted (delignified) agricultural materials

The strain SG6 at 50 and 100 g AECS, AEPS, AESS and alkali-extracted grass (AEG) l^{-1} efficiently degraded 81, 82, 79, 77 and 70, 71, 68, 67% of substrates, respectively. A maximum of 21.12 g acetic acid/41.2 g substrate utilized at 50 g AEPS l^{-1} and 30.98 g acetic acid/70.6 g substrate utilized at 100 g AEPS l^{-1} was obtained. The acetic acid yields of the strain at 50 g l^{-1} and 100 g substrates l^{-1} were more or less similar to the values obtained with pure cellulose (Whatman No.1 filter paper) (Tables 4 & 5). At 100 g substrate l^{-1}, the acetic acid yield of the strain decreased considerably with an increase in ethanol production and reducing sugars accumulation. In general AEPS and AECS served as the best substrates for acetic acid production by the strain SG6, followed by AESS and AEG.

The significant interactions were observed among the substrates and between substrate and its concentrations for the production of acetic acid. There is also significantly increased acetic acid production observed with the increasing concentration of 50 to 100 g substrate l^{-1}. However, no significant interaction was observed between cotton straw and paddy straw, within concentration of 50 and 100 g l^{-1} (Table 5).

Discussion

The ability of the *C. lentocellum* to grow on and degrade pure cellulose and alkali-treated agricultural materials efficiently indicates the presence of a true cellulase (Duong *et al.* 1983; Beguin & Aubert 1994) and a hemicellulase system (Gilbert & Hazel wood 1993) (Tables 1 & 3). Feeble growth on carboxymethylcellulose sodium salt indicates that the substitution of glucose units by carboxymethyl groups resulted in their poor utilization. Freier *et al.* (1988) in *C. thermocellum* JW20 and Rasmussen *et al.* (1988) in *Ruminococcus flavifaciens* FD1 have observed that growth of the organism was dependent on the extent of the substitution, the higher the substitution the lesser the growth. Sai Ram *et al.* (1991) reported decreased yields of ethanol and acetic acid in *C. thermocellum* SS8 and GS1 in carboxymethylcellulose compared to other pure cellulosic substrates.

The poor growth of strain SG6 on crude biopolymers may be because of the presence of soluble inhibitors that were removed after hot water treatment resulting in appreciable growth and substrate degradation (Table 2). Alkali treatment further delignified the substrates, rendering them more susceptible to degradation (Table 3) (Donefer *et al.* 1969; Datta 1981; Kundu *et al.* 1983; Sai Ram & Seenayya 1991). The acetate to ethanol ratio of the fermentation by *C. lentocellum* SG6 is towards ethanol with pretreated biomass as substrate compared to pure cellulosic substrates (Tables 1–3). This characteristic of acetogenic *C. lentocellum* SG6 is opposite to that of ethanologenic organisms such *C. thermocellum*, which show a shift away from ethanol and towards acetate when grown on biomass materials (Sai Ram & Seenayya 1991).

The fermentation of pure cellulosics and treated agricultural materials and production of acetic acid by *C. lentocellum* SG6 is more efficient than other reported monoculture, cellulolytic, bacterial strains (Fond *et al.* 1983; Khan *et al.* 1984; Ruyet *et al.* 1984; Sai Ram & Seenayya 1991). Fond *et al.* (1983) reported low acetic acid yields by *Clostridium* sp. H10 at 6 g Solka Floc l^{-1}

Table 5. Fermentation of higher concentrations of alkali-extracted agricultural materials by *C. lentocellum* SG6.

Substrate concentration (g l^{-1})	Acetic acid (g l^{-1})	Ethanol (g l^{-1})	Reducing sugars (g l^{-1})	Substrate degraded (g l^{-1})	Acetate yield (g g^{-1})	A/E ratio
Substrate vs. concentration						
50^{x}						
Cotton straw	21.05[a]	4.86[a]	0.53[a]	40.4[a]	0.52[a]	4.33[a]
Paddy straw	21.12[a]	4.93[a,c]	0.96[b]	41.2[b]	0.51[b]	4.28[a]
Sorghum stover	19.43[b]	4.60[b]	0.72[c]	39.4[c]	0.49[c]	4.22[b]
Grass	18.71[c]	4.95[c]	0.41[d]	38.6[d]	0.48[d]	3.78[c]
100^{xx}						
Cotton straw	30.86[d]	8.78[d]	0.92[e]	70.1[e]	0.44[e]	3.51[d]
Paddy straw	30.98[d]	8.72[d]	1.51[f]	70.6[e]	0.44[e]	3.55[d]
Sorghum stover	28.35[e]	7.22[e]	1.16[g]	68.4[f]	0.41[f]	3.93[e]
Grass	27.05[f]	7.90[e]	0.78[h]	67.5[g]	0.40[g]	3.42[f]
C.D. at 5%	0.357	0.074	0.023	0.649	0.006	0.051
F-value	22.35*	293.35*	45.99*	1.31*	4.02*	99.03*
Concentration: (Mean values of all substrates at 50 and 100 g l^{-1} concentration)						
50	20.08[a]	4.84[a]	0.66[a]	39.9[a]	0.50[a]	4.16[a]
100	29.39[b]	8.16[b]	1.09[b]	69.2[b]	0.42[b]	3.61[b]
C.D. at 5%	0.178	0.037	0.012	0.325	0.003	0.026
F-value	10955.96*	32118.04*	5758.25*	32584.25*	2392.82*	1803.30*
Substrates: (Mean values of 50 and 100 g substrate l^{-1} concentration)						
Cotton straw	25.96[a]	6.82[a]	0.72[a]	55.3[a]	0.48[a]	3.92[a]
Paddy straw	26.22[b]	6.82[a]	1.24[b]	55.9[b]	0.48[a]	3.94[a]
Sorghum stover	23.89[c]	5.91[b]	0.94[c]	53.9[c]	0.45[b]	4.08[b]
Grass	22.88[d]	6.43[c]	0.59[d]	53.1[d]	0.44[c]	3.60[c]
C.D. at 5%	0.252	0.052	0.016	0.459	0.005	0.036
F-value	329.82*	545.52*	2385.96*	63.26*	134.38*	247.83*

* $P < 0.0001$.

The values are the average of three experiments, each in triplicate.

Two way ANOVA with interactions was utilized; Different superscripts indicate that they are significantly different from one another at $P < 0.05$ level in interaction of substrate vs. concentrations of 50 and 100 g l^{-1} and substrates.

[x] Incubated for 9 days; [xx] incubated for 14 days.

g g^{-1}: gram acetic acid per gram substrate; A/E: Acetic acid to ethanol ratio.

$CaCO_3$ was used at 50% of the substrate concentration as buffering agent.

with 65% substrate degradation. Ruyet *et al.* (1984) reported 7.17 g (119.5 mM) acetic acid in monoculture fermentation by *C. thermocellum* TC11 and 16.2 g (270 mM) acetic acid in coculture fermentation by *C. thermocellum* and *Acetogenium kivui* from 18 g (100 mM) glucose equivalents fermented. Miller & Wolin (1995) reported 6.54 g (109 mM) acetic acid from 9.36 g (52 mM) hexose equivalents fermented in coculture fermentation by *Ruminococcus albus* and a hydrogen utilizing acetogen.

The acetic acid production by *C. lentocellum* SG6 is also encouraging at higher substrate concentrations using $CaCO_3$ as buffering agent in fermentation medium. Such a fermentation process may also result in the formation of calcium acetate as a product. The calcium acetate can be used as large volume chemical for industrial applications (Wise *et al.* 1991; Cheryan *et al.* 1997). Calcium acetate, calcium magnesium acetate and other acetate salts are reported to be used in large scale applications for deicing of roads (Wise *et al.* 1991; Wise 1992), in heat-exchange fluid, eliminating sulphur in coal mines etc. (Levendis 1991; Manivanan & Wise 1991).

Acetic acid production of 0.67 g g^{-1} of pure cellulose (Whatman No. 1 filter paper) and 0.63 g g^{-1} of alkali treated cotton straw by *C. lentocellum* SG6 are the highest in monoculture fermentations by cellulolytic bacteria reported so far. Therefore, *C. lentocellum* SG6 has considerable potential as an industrial strain for the direct conversion of cellulosic material to acetic acid. By utilizing this organism as production strain, acetic acid can be produced by fermentation using abundantly available cheap cellulosic biomass as substrates. The direct fermentation of cellulose to acetic acid is a significant process to avoid the conventional multi-step fermentative production of acetic acid.

Acknowledgement

The authors thank Dr K. Visweswara Rao (National Institute of Nutrition (NIN), Hyderabad, India) for help with the statistical analysis.

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